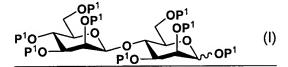
AMENDMENTS TO THE CLAIMS

- 1. (Cancelled)
- 2. (Currently amended) The A method for preparing a trisaccharide (Manβ1→4GlcNβ1→4GlcN) of the a reducing terminal in the a core sugar chain structure of an asparagine-linked glycoprotein, of claim 1, further comprising each of

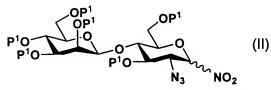
(1) a process of preparing a mannose disaccharide compound (a type of ManP¹β1→4ManP¹) of the formula (I)



wherein P¹ is an OH-protecting group selected from the group consisting of acetyl, benzyl, 4-methoxybenzyl, benzoyl, methoxymethyl, tetrahydropyranyl, trimethylsillyl and triethyl silyl, and the wavy line means that -OP¹ is linked at an axial or equatorial position, or mixture of both,

by hydrolyzing a polysaccharide having mannose β -1,4-bonds and protecting OH groups of the resulting hydrolysate,

- (2) a process of preparing a glycal compound, in which mannose of the <u>a</u> reducing terminal of the mannose disaccharide is converted to glycal, by halogenation and reduction of the mannose disaccharide (a type of ManP¹β1→4ManP¹), and
- (3) a process of preparing an azide disaccharide compound (a type of ManP¹β1→4ManP¹) shown with formula (II) in which the a_2-azide group of mannose in the a_reducing terminal is linked at the an equatorial position;



wherein P¹ is the same <u>as described</u> above, the wavy line means that -NO₂ is linked at an axial or equatorial position, or mixture of both,

by azidenitration reaction of the glycal compound above.

- 3. (Currently amended) The method for preparing a trisaccharide (Manβ1→4GlcNβ1→4GlcN) of the <u>a</u> reducing terminal in the <u>a</u> core sugar chain structure of an asparagine-linked glycoprotein of claim 2, further comprising
- (4) a process of substituting the nitro group of the azide disaccharide compound (a type of ManP¹β1→4ManP¹) with a leaving group selected from the group consisting of fluorine atom, chlorine atom, trihaloacetoimidate, pentenyl, alkylthio and arylthio, and
- (5) a process of preparing a trisaccharide compound (a type of Man β 1 \rightarrow 4GlcNP¹ β 1 \rightarrow 4GlcNP²) shown with the formula (III);

$$P_{10}^{10} \longrightarrow P_{10}^{10} \longrightarrow$$

wherein P¹, P², P³ and P¹¹ are is an OH- protecting group, as described above, P² is an OH-protecting group selected from the group consisting of acetyl, benzyl, 4-methoxybenzyl, benzoyl, methoxymethyl, tetrahydropyranyl, trimethylsillyl and triethyl silyl, P³ is an amino-protecting group selected from the group consisting of phthalimide, tert-butyloxycarbonyl, benzyloxycarbonyl, acetyl, benzoyl and benzyl, and P¹¹ is an OH-protecting group selected from the group consisting of acetyl, benzyl, 4-methoxybenzyl, benzoyl, methoxymethyl, tetrahydropyranyl, trimethylsillyl and triethyl silyl, the same above.

by a reaction of the product having the leaving group with amino-protected glucopyranoside shown with the formula;

wherein P², P³ and P¹¹ are the same as described above. is an OH-protecting group, P³ is an amino-protecting group and P¹¹ is an OH protecting group.

- 4. (Currently amended) The method for preparing a trisaccharide (Manβ1→4GlcNβ1→4GlcN) of the a_reducing terminal in the a_core sugar chain structure of an asparagine-linked glycoprotein of claim 3, further comprising
- (6) a process of preparing an asparagine-linked trisaccharide (Manβ1→4GlcNP¹β1→4GlcNP²) compound shown with the formula (IV);

wherein P¹ and P² are <u>independently OH-protecting groups</u>, as described above the same above, P⁴ and P⁶ are independently amino-protecting groups <u>selected from the group consisting of phthalimide</u>, tert-butyloxycarbonyl, benzyloxycarbonyl, acetyl, benzoyl and <u>benzyl</u>, and P⁵ is a carboxyl-protecting group <u>which is t-Bu</u>, by deprotecting the P¹¹ group of the compound (III),

$$P_{10}^{10} = 0$$
 $P_{10}^{10} = 0$
 P_{10}^{10

wherein P^1 , P^2 and P^{11} are independently OH-protecting groups, as described above, and P^3 is an amino-protecting group, as described above,

reducing the azide group to an amino group, protecting the amino group with an acetyl group, forming an oxazoline ring simultaneously with deprotecting a hydroxy group of a reducing terminal, and coupling with a protected asparagines derivative after introducing a -N=C=S group at the reducing terminal coupling of the reducing terminal of the trisaccharide compound above with the protected asparagine derivative.

5. (Currently amended) A method for preparing a mannose disaccharide compound

(a type of $ManP^1\beta 1 \rightarrow 4ManP^1$) shown with the formula (I);

$$P^{1}O$$
 $P^{1}O$
 $P^{1}O$
 $P^{1}O$
 $P^{1}O$
 $P^{1}O$
 $OP^{1}O$
 $OP^{1}O$

wherein P¹ is an OH-protecting group <u>selected from the group consisting of acetyl</u>, <u>benzyl</u>, <u>4-methoxybenzyl</u>, <u>benzoyl</u>, <u>methoxymethyl</u>, <u>tetrahydropyranyl</u>, <u>trimethylsillyl</u> and <u>triethyl silyl</u>, and the wavy line means that -OP¹ is linked at an axial or equatorial position, or mixture of both,

by hydrolyzing guar gum or galactomannan of the formula (V);

wherein n is an integer of 50 or more, a polysaccharide having mannose β -1,4-bonds and protecting OH groups of the resulting hydrolysate.

6. (Currently amended) A method for preparing the-an azide disaccharide (a type of ManP¹β1→4ManP¹) shown with the formula (II) in which the-a 2-azide group of mannose in the-a reducing terminal is linked at the-an equatorial position;

$$P^{1}O$$
 $P^{1}O$
 $P^{1}O$
 OP^{1}
 O

wherein P¹ is an OH-protecting group <u>selected from the group consisting of acetyl</u>, <u>benzyl</u>, <u>4-methoxybenzyl</u>, <u>benzoyl</u>, <u>methoxymethyl</u>, <u>tetrahydropyranyl</u>, <u>trimethylsillyl</u> and <u>triethyl silyl</u>, and the wavy line means that –NO₂ is linked at an axial or equatorial position, or mixture of both,

comprising a process of preparing a glycal compound, in which mannose of the reducing

Shinichiro NISHIMURA et al. Attorney Docket No. 2006_0977A Serial No. 10/584,065 March 26, 2008

terminal of the mannose disaccharide is converted to glycal, by halogenation and reduction of the mannose disaccharide compound (a type of ManP¹ β 1 \rightarrow 4ManP¹) shown with the formula (I);

$$P^{1}O P^{1} O P^{1}$$

wherein P¹ is the same <u>as described</u> above and the wavy line means that -OP¹ is linked at an axial or equatorial position, or mixture of both, and subsequent azidenitration reaction of the glycal compound.

7. (Currently amended) A method for preparing the <u>a</u>trisaccharide compound shown with the formula (III);

wherein P¹, P² and P¹¹ are independently OH- protecting groups selected from the group consisting of acetyl, benzyl, 4-methoxybenzyl, benzoyl, methoxymethyl, tetrahydropyranyl, trimethylsillyl and triethyl silyl, and P³ is an amino-protecting group selected from the group consisting of phthalimide, tert-butyloxycarbonyl, benzyloxycarbonyl, acetyl, benzoyl and benzyl, wherein P¹, P², P³ and P¹¹ are the same above;

comprising a process of substituting the nitro group of the azide disaccharide compound (a type of ManP¹β1→4ManP¹) shown with the formula (II) with a leaving group selected from the group consisting of fluorine atom, chlorine atom, trihaloacetoimidate, pentenyl, alkylthio and arylthio;

$$P^{1}O \longrightarrow P^{1}O \longrightarrow P^{1}O \longrightarrow N_{3} \longrightarrow NO_{2}$$
 (II)

wherein P¹ is the same <u>as described</u> above, the wavy line means that -NO₂ is linked at an axial or equatorial position, or mixture of both, and the <u>a</u> 2-azide group of mannose in the reducing terminal is linked at the equatorial position,

and next, reacting the substituted compound having the leaving group with aminoprotected glucopyranoside of the formula;

wherein P², P³ and P¹¹ are is an OH-protecting group the same as described above. , P³ is an amino-protecting group and P¹¹ is an OH-protecting group.

8. (Currently amended) A method for preparing an asparagine-linked trisaccharide compound (Man β 1 \rightarrow 4GlcNP¹ β 1 \rightarrow 4GlcNP²) shown with the formula (IV)

wherein P¹ and P² are <u>independently OH- protecting groups selected from the group consisting of acetyl, benzyl, 4-methoxybenzyl, benzoyl, methoxymethyl, tetrahydropyranyl, trimethylsillyl and triethyl silyl, the same above, P⁴ and P⁶ are independently amino-protecting groups <u>selected from the group consisting of phthalimide, tert-butyloxycarbonyl, benzyloxycarbonyl, acetyl, benzoyl and benzyl, and P⁵ is a carboxyl-protecting group <u>which is t-Bu</u>, by coupling of the reducing terminal of the trisaccharide <u>deprotecting the P¹¹ group of the compound (III)</u>,</u></u>

$$P_{10}^{10} \xrightarrow{OP_{1}^{1}} OP_{10}^{10} \xrightarrow{OP_{10}^{1}} OP_{10}^{10} OP_{10}^{10} OP_{10}^{10} OP_{11}^{10} O$$

wherein P¹, and P² are the same as described above, P³ is an amino-protecting group selected from the group consisting of phthalimide, tert-butyloxycarbonyl, benzyloxycarbonyl, acetyl, benzoyl and benzyl, and P¹¹ is an OH-protecting group selected from the group consisting of acetyl, benzyl, 4-methoxybenzyl, benzoyl, methoxymethyl, tetrahydropyranyl, trimethylsillyl and triethyl silyl, reducing the azide group to an amino group, protecting the amino group with an acetyl group, forming an oxazoline ring simultaneously with deprotecting a hydroxy group of a reducing terminal, and coupling with a protected asparagines derivative after introducing a -N=C=S group at the reducing terminal. and P¹¹ are the same above, with a protected asparagine derivative.

9. (Currently amended) The An azide disaccharide (a type of ManP¹ β 1 \rightarrow 4ManP¹) compound shown with the formula (II);

$$P_{10}^{10} \xrightarrow{OP_{1}^{1}} OP_{10}^{10} \xrightarrow{OP_{10}^{1}} OP_{10}^{10} O$$

wherein P¹ is an OH-protecting group selected from the group consisting of acetyl, benzyl, 4-methoxybenzyl, benzoyl, methoxymethyl, tetrahydropyranyl, trimethylsillyl and triethyl silyl, and the wavy line means that -NO₂ is linked at an axial or equatorial position, or mixture of both.

10. (Currently amended) The A trisaccharide compound (a type of Man β 1 \rightarrow 4GlcNP¹ β 1 \rightarrow 4GlcNP²) shown with the formula of (III);

$$P_{P_{1}O}^{10} = P_{1}^{10} = P_{1}^{10}$$

wherein P¹, P² and P¹¹ are <u>independently OH-protecting groups selected from the group consisting of acetyl, benzyl, 4-methoxybenzyl, benzoyl, methoxymethyl, tetrahydropyranyl, trimethylsillyl and triethyl silyl, and P³ is an amino-protecting group selected from the group consisting of phthalimide, tert-butyloxycarbonyl, benzyloxycarbonyl, acetyl, benzoyl and benzyl.</u>

- 11. (New) A method for preparing a trisaccharide (Manβ1→4GlcNβ1→4GlcN) of a reducing terminal in a core sugar chain structure of an asparagine-linked glycoprotein, comprising
- (1) a process of preparing a mannose disaccharide compound (a type of $ManP^1\beta 1 \rightarrow 4ManP^1$) of the formula (I)

$$P^{1}O$$
 $P^{1}O$
 P

wherein P¹ is an OH-protecting group selected from the group consisting of acetyl, benzyl, 4-methoxybenzyl, benzoyl, methoxymethyl, tetrahydropyranyl, trimethylsillyl and triethyl silyl, and the wavy line means that -OP¹ is linked at an axial or equatorial position, or mixture of both,

by hydrolyzing guar gum or galactomannan of the formula (V);

Shinichiro NISHIMURA et al. Attorney Docket No. 2006_0977A Serial No. 10/584,065 March 26, 2008

wherein n is an integer of 50 or more, and protecting OH groups of the resulting hydrolysate.

- 12. (New) The method for preparing a trisaccharide (Man β 1 \rightarrow 4GlcN β 1 \rightarrow 4GlcN) of a reducing terminal in a core sugar chain structure of an asparagine-linked glycoprotein of claim 11, further comprising each of
- (2) a process of preparing a glycal compound, in which mannose of a reducing terminal of the mannose disaccharide is converted to glycal, by halogenation and reduction of the mannose disaccharide (a type of $ManP^1\beta 1 \rightarrow 4ManP^1$), and
- (3) a process of preparing an azide disaccharide compound (a type of ManP¹β1→4ManP¹) shown with formula (II) in which a 2-azide group of mannose in a reducing terminal is linked at an equatorial position;

$$P_{10}^{10} \xrightarrow{OP_{1}^{1}} OP_{10}^{10} \xrightarrow{OP_{10}^{1}} OP_{10}^{10} O$$

wherein P¹ is the same as described above, the wavy line means that -NO₂ is linked at an axial or equatorial position, or mixture of both, by azidenitration reaction of the glycal compound above.

- 13. (New) The method for preparing a trisaccharide (Manβ1→4GlcNβ1→4GlcN) of a reducing terminal in a core sugar chain structure of an asparagine-linked glycoprotein of claim 12, further comprising
- (4) a process of substituting the nitro group of the azide disaccharide compound (a type of ManP¹β1→4ManP¹) with a leaving group selected from the group consisting of fluorine atom, chlorine atom, trihaloacetoimidate, pentenyl, alkylthio and arylthio, and

(5) a process of preparing a trisaccharide compound (a type of $Man\beta1 \rightarrow 4GlcNP^1\beta1 \rightarrow 4GlcNP^2$) shown with the formula (III);

$$P_{P_{10}}^{10} \xrightarrow{OP_{1}} OP_{P_{10}}^{10} OP_{N_{3}}^{10} OP_{N_{20}}^{10} OP_{N_{10}}^{10} OP_{N_{10}}^{$$

wherein P¹ is an OH- protecting group, as described above, P² is an OH-protecting group selected from the group consisting of acetyl, benzyl, 4-methoxybenzyl, benzoyl, methoxymethyl, tetrahydropyranyl, trimethylsillyl and triethyl silyl, P³ is an aminoprotecting group selected from the group consisting of phthalimide, tert-butyloxycarbonyl, benzyloxycarbonyl, acetyl, benzoyl and benzyl, and P¹¹ is an OH-protecting group selected from the group consisting of acetyl, benzyl, 4-methoxybenzyl, benzoyl, methoxymethyl, tetrahydropyranyl, trimethylsillyl and triethyl silyl, by a reaction of the product having the leaving group with amino-protected glucopyranoside shown with the formula;

wherein P², P³, and P¹¹ are the same as described above.

- 14. (New) The method for preparing a trisaccharide (Manβ1→4GlcNβ1→4GlcN) of a reducing terminal in a core sugar chain structure of an asparagine-linked glycoprotein of claim 13, further comprising
- (6) a process of preparing an asparagine-linked trisaccharide $(Man\beta1 {\rightarrow} 4GlcNP^1\beta1 {\rightarrow} 4GlcNP^2) \ compound \ shown \ with \ the \ formula \ (IV);$

Shinichiro NISHIMURA et al. Attorney Docket No. 2006_0977A Serial No. 10/584,065 March 26, 2008

wherein P¹ and P² are independently OH- protecting groups, as described above, P⁴ and P⁶ are independently amino-protecting groups selected from the group consisting of phthalimide, tert-butyloxycarbonyl, benzyloxycarbonyl, acetyl, benzoyl and benzyl, and P⁵ is a carboxyl-protecting group which is t-Bu,

by deprotecting the P¹¹ group of the compound (III),

$$P_{P_{10}}^{10} \xrightarrow{OP_{1}} OP_{N_{3}}^{10} OP_{N_{9}}^{20} OP_{N_{11}}^{20} OP_{N_{11}}^{2$$

wherein P^1 , P^2 and P^{11} are independently OH- protecting groups, as described above, and P^3 is an amino-protecting group, as described above,

reducing the azide group to an amino group, protecting the amino group with an acetyl group, forming an oxazoline ring simultaneously with deprotecting a hydroxy group of a reducing terminal, and coupling with a protected asparagine derivative after introducing a -N=C=S group at the reducing terminal.